DECLARATION UNDER 37 C.F.R. § 1.132		
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S I R:

I, Catherine Amiel, declare that:

1. I am a co-inventor of the present application. I have been involved in all stages of the development of the present invention and during the patent application process. I am aware of the contents of pending claims. Further, I have reviewed the Office Action mailed September 4, 2008 and the prior art cited in the Office Action.

2. The present invention is directed to a composition comprising an aqueous dispersion of particles (p) of mean diameter between 80 and 5000 nm. See, e.g., claim 1. The particles (p) contain compounds (A) and (B). Particles (p) comprising compounds (A) and (B) are not water-soluble. However, compounds (A) and (B), in an isolated state, are water soluble. The aqueous medium of the composition, which comprises the claimed aqueous dispersion, may contain dissolved compounds (A) and (B), in addition to the discrete particles (p), which are not dissolved in the aqueous medium.

3. In varying forms of the invention, 80% of a total mass of (A) and (B) in the composition may be contained in the particles (p). For example such a composition is

recited in claim 11. In the case that at least 80% of the total mass of compounds (A) and (B) are contained in particles, the remaining 20% or less of the mass of compounds (A) and (B) will be dissolved in the aqueous solution. This property is disclosed in the present specification, page 14, third paragraph, which describes that the majority of the polymers (A) and the macromolecules (B) are localized in the particles (p) and thus, in general, at least 80% of (A) and (B), by mass, present in the composition are contained in the particles (p). Claim 11 has been amended to now more clearly recite that the composition may contain compounds (A) and (B) dissolved in the aqueous solution comprising the aqueous dispersion of particles (p), wherein at least 80% by mass of the compounds (A) and (B) present in the composition are contained in the particles (p).

4. The compounds (B) macromolecules of polysaccharides comprise at least three groups G. The groups G are capable of forming inclusion complexes with the cyclodextrins present in the structure of the polymers (A). See, e.g., claim 1. The recited term "groups G" is a term in the art that would be understood by one of ordinary skill in the art at the time of the invention to mean groups capable of forming inclusion complexes with the cyclodextrins. Attached in Appendix A to this Declaration is an article entitled *Pharmaceutical Applications of Cyclodextrins. 1. Drug Solubilization and Stabilization*, Loftsson and Brewster, appearing in the Journal of Pharmaceutical Sciences, October 1996 (hereinafter "Loftsson"). Loftsson provides evidence that one of ordinary skill in the art would have understood at the time of the invention what the groups G were.

5. Further, the present specification provides a sufficient disclosure to allow one of ordinary skill in the art to know which groups G are capable of forming inclusion complexes. Accordingly, one skilled in the art would be able to practice the invention as claimed.

6. In one particular form the groups G are aliphatic groups, linear or branched having 8 to 18 carbon atoms. For example, see claim 7. One further aliphatic group is a C_{12} aliphatic group, i.e., one which has 12 carbon atoms. Again, claim 7 which recites that the aliphatic group has between 8 and 18 carbon atoms covers the form in which the aliphatic group contains 12 carbon atoms.

7. The macromolecules of polysaccharides comprising groups G are capable of forming inclusion complexes. One skilled in the art would understand that the term "capable" means that the macromolecules may, but not necessarily, will form complexes with the cyclodextrins present in the structure. The reason that the groups may not necessarily form complexes is due to not being able to guarantee that all macromolecules are complexed due to variabilities in the particle (p) structure and the exact form of compound (A). The presence of complexes of cyclodextrins and macromolecules in the claimed composition is compulsory. However, it is possible that all of the macromolecules are not complexed, e.g., one of the cases being when there is not enough cyclodextrins in the composition, although the free macromolecules comprising groups G are nevertheless still capable for forming inclusion complexes.

8. I am a co-inventor and thus very familiar with the ACS Symposium reference *Stimuli-Responsive Water Soluble and Amphiphilic Polymers* "Macromolecular Assemblies Generated by Inclusion Complexes Between Amphiphilic

Polymers and β-Cyclodextrin Polymers in Aqueous Media", Amiel et al (hereinafter "Amiel") cited in the Office Action of September 4, 2008. The present composition is distinguishable from the compositions disclosed in Amiel. The Amiel reference teaches that a mixture Dextran-Adamantan with β-cyclodextrin/epichlorohydrin oligomers : 50/50 (w/w) leads to aggregates (page 71). Further, Amiel Figure 11 shows that the hydrodynamic radius of the aggregates depends in the adamantan concentration. The radius indicated in Figure 11 is lower than 30 nm, and is therefore lower than the radius of the particles contained in the as now claimed composition (i.e. particles with a radius between 40 and 2500 nm (diameter between 80 to 5000 nm). Thus, the particles of the present invention differ from Amiel's aggregate in that the present particles are larger.

9. In the outstanding Office Action, the Examiner alleges that, from Amiel, one of ordinary skill in the art would be able to obtain bigger aggregates corresponding to particles claimed. However, the present composition differs from the aggregates disclosed in Amiel. One notable difference is the stability. For example, the claimed particles provide for a thermodynamically stable system (see present specification, page 4, first full paragraph). As described thoroughly in the present specification, the storage and dilution of the claimed composition is possible since the composition is stable. Moreover, the stability of the composition is a direct result of the claimed particles which form the aqueous dispersion.

10. Nowhere in Amiel is there any teaching, let alone anything to lead one of ordinary skill in the art, to believe enhanced stability of its aggregate could be achieved by modifying its disclosure. To the contrary, the Amiel disclosure specifically relates to a mixture comprising aggregates which do not present the stability which inherently

flows from the claimed invention as described in the present specification. Moreover, Amiel fails to provide an enabling disclosure to allow one of ordinary skill in the art to form an aqueous dispersion with particles having the sizes claimed.

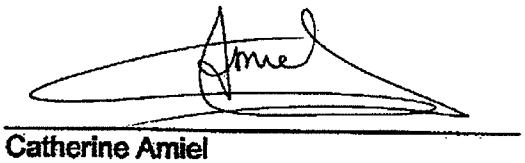
11. Further, there fails to be any disclosure in Amiel to lead one to form larger particles. Amiel is specifically directed to forming an aggregate with a specific size. Moreover, it would not have been obvious to one of ordinary skill in the art to modify the composition of Amiel to have the claimed size. Amiel is directed to a very specific complex of polymers with β-cyclodextrin polymers in an aqueous media. In particular, the reference is specifically directed to an aqueous solution and not a dispersion as claimed. Furthermore, it would be contrary to its teaching, which is directed to a solution of polymers, to modify the constituents to result in a dispersion of particles having the claimed size since Amiel is specifically directed to a solution with specific properties as disclosed. More importantly, Amiel is specifically directed to forming an aqueous solution of amphiphilic polymers and thus it is important that a particular solution be formed. Conversely, the present invention is directed to a dispersion and thus a completely different aqueous composition. Accordingly, one of ordinary skill in the art would not modify the composition of Amiel to form a dispersion having the particle size as claimed as doing so would thwart the teaching of Amiel.

12. In order to demonstrate that the present composition is different than the aggregate of Amiel, the following experiment was conducted. A centrifugation test at 5000 g for one hour in which a test tube containing either the claimed composition or the Amiel aggregate was spun. After the centrifugation test, a centrifugation pellet was observed at the bottom of the test tube containing the present particles. The centrifuge

pellet formed because of the high density of the present particles. However, after centrifugation of the test tube containing the aggregate of Amiel, no pellet was observed. Accordingly, the claimed aqueous dispersion is distinguishable from the Amiel aggregate.

13. The undersigned declares further that all statements made herein of his knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under section 1001 of Title 18 of United States Code and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Signed this 2nd day of March 2009.



Catherine Amiel

APPENDIX A

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REVIEW ARTICLE

Pharmaceutical Applications of Cyclodextrins. 1. Drug Solubilization and Stabilization

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Abstract □ Cyclodextrins are cyclic oligosaccharides which have recently been recognized as useful pharmaceutical excipients. The molecular structure of these glucose derivatives, which approximates a truncated cone or torus, generates a hydrophilic exterior surface and a nonpolar cavity interior. As such, cyclodextrins can interact with appropriately sized molecules to result in the formation of inclusion complexes. These noncovalent complexes offer a variety of physicochemical advantages over the unmanipulated drugs including the possibility for increased water solubility and solution stability. Further, chemical modification to the parent cyclodextrin can result in an increase in the extent of drug complexation and interaction. In this short review, the effects of substitution on various cyclodextrin properties and the forces involved in the drug–cyclodextrin complex formation are discussed. Some general observations are made predicting drug solubilization by cyclodextrins. In addition, methods which are useful in the optimization of complexation efficacy are reviewed. Finally, the stabilizing/destabilizing effects of cyclodextrins on chemically labile drugs are evaluated.

Introduction

Although cyclodextrins are frequently regarded as a new group of pharmaceutical excipients, they have been known for over 100 years.[1] The foundations of cyclodextrin chemistry were laid down in the first part of this century[2,3] and the first patent on cyclodextrins and their complexes was registered in 1953.[4] However, until 1970 only small amounts of cyclodextrins could be produced and high production costs prevented their widespread usage in pharmaceutical formulations. Recent biotechnological advancements have resulted in dramatic improvements in cyclodextrin production, which has lowered their production costs. This has led to the availability of highly purified cyclodextrins and cyclodextrin derivatives which are well suited as pharmaceutical excipients. These carbohydrates are mainly used to increas aqueous solubility, stability, and bioavailability of drugs they can also, for example, be used to convert liquid d into microcrystalline powders, prevent drug–drug or d additive interactions, reduce gastrointestinal or ocular i tion, and reduce or eliminate unpleasant taste and sme

The following is a short review of the effects of cyclodex on the solubility and stability of drugs in aqueous solu with emphasis on the more recent developments. For fu information on cyclodextrins and their physicochemical erties the reader is referred to several excellent books reviews published in recent years.[5–13]

Structure and Physicochemical Properties

Cyclodextrins are cyclic (α-1,4)-linked oligosaccharid α-D-glucopyranose containing a relatively hydrophobic ce cavity and hydrophilic outer surface. Owing to lack of rotation about the bonds connecting the glucopyranose u the cyclodextrins are not perfectly cylindrical molecule: are toroidal or cone shaped. Based on this architecture primary hydroxyl groups are located on the narrow si the torus while the secondary hydroxyl groups are locat the wider edge (Figure 1). The most common cyclodex are α-cyclodextrin, β-cyclodextrin, and γ-cyclodextrin, v consist of six, seven, and eight glucopyranose units, re tively. While it is thought that, due to steric factors, dextrins having fewer than six glucopyranose units ca exist, cyclodextrins containing nine, ten, eleven, twelve thirteen glucopyranose units, which are designated δ-, η-, and θ-cyclodextrin, respectively, have been reporte Of these large-ring cyclodextrins only δ-cyclodextrin has well characterized.[16,17] Chemical and physical properti the four most common cyclodextrins are given in Table 1 melting points of α-, β-, and γ-cyclodextrin are betwee and 265 °C, consistent with their stable crystal l structure.[18]

The parent cyclodextrins, in particular β-cyclodextrin, limited aqueous solubility, and their complex formation

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Table 2—Some Currently Available Cyclodextrins Obtained by Substitution of the OH Groups Located on the Edge of the Cyclodextrin Ring[a]

Cyclodextrin Derivatives		
α	β	γ
	Alkylated:	
Methyl	Methyl	Methyl
	Ethyl	
Butyl	Butyl	Butyl
		Pentyl
	Hydroxylalkylated:	
	Hydroxyethyl	Hydroxyethyl
2-Hydroxypropyl	2-Hydroxypropyl	2-Hydroxypropyl
	2-Hydroxybutyl	
	Esterified:	
Acetyl	Acetyl	Acetyl
	Propionyl	
	Butyryl	
Succinyl	Succinyl	Succinyl
	Benzoyl	
	Palmityl	
	Toluenesulfonyl	
	Esterified and Alkylated:	
	Acetyl methyl	
	Acetyl butyl	
	Branched:	
Glucosyl	Glucosyl	Glucosyl
Maltosyl	Maltosyl	Maltosyl
	Ionic:	
Carboxymethyl ether	Carboxymethyl ether	Carboxymethyl ether
	Carboxymethyl ethyl	
Phosphate ester	Phosphate ester	Phosphate ester
	3-Trimethylammonium-2-hydroxypropyl ether	
	Sulfobutyl ether	
	Polymerized:	
Simple polymers	Simple polymers	Simple polymers
Carboxymethyl	Carboxymethyl	Carboxymethyl

[a] Since both the number of substitutes and their location will affect the physicochemical properties of the cyclodextrin molecules, such as their aqueous solubility and complexing abilities, each derivative listed should be regarded as a group of closely related cyclodextrin derivatives.

graphic retention times.[40] While it is possible to use both guest or host changes to generate equilibrium constants, guest properties are usually most easily assessed. Connors has evaluated the population characteristics of cyclodextrin complex stabilities in aqueous solution.[41]

The thermodynamic parameters, i.e., the standard free energy change (ΔG), the standard enthalpy change (ΔH), and the standard entropy change (ΔS), can be obtained from the temperature dependence of the stability constant of the cyclodextrin complex.[42] The thermodynamic parameters for several series of drugs and other compounds have been determined and analyzed.[43–45] The thermodynamic parameters of several other drugs are listed in Table 3. The complex formation is almost always associated with a relatively large negative ΔH and a ΔS that can be either positive or negative. Also, complex formation is largely independent of the chemical properties of the guest (i.e., drug) molecules. The association of binding constants with substrate polarizability suggests that van der Waals forces are important in complex formation.[50] Hydrophobic interactions are associated with a slightly positive ΔH and a large positive ΔS; therefore, classical hydrophobic interactions are entropy driven, suggesting that they are not involved with cyclodextrin complexation since, as indicated, these are enthalpically driven processes. Furthermore, for a series of guests there tends to be a linear relationship between enthalpy and entropy, with increasing

Table 3—Standard Enthalpy Change (ΔH) and Standard Entropy Ch (ΔS) for Several Drug–Cyclodextrin Complexes

Cyclodextrin[a]	Drug	pH	ΔH (kJ/mol)	ΔS (J/(mol K))
HP-α-CD	Hydrocortisone		−32	−70
β-CD	Phenytoin, un-ionized	7	−38	−67
	Phenytoin, ionized	7	−21	−21
β-CD	Naproxen		−13	18
β-CD	Adenine arabinoside	7	−28	−64
β-CD	Adenosine	7	−21	−53
β-CD	Ibuprofen (pK_a 5.2)	2	−29	15
		4	−32	4
		5	−29	3
		6	−17	34
β-CD	Diazepam (pK_a 3.3)	2	−0.2	70
		3	−3.3	69
		4	−17	22
		6	−18	19
β-CD	Hydrochlorothiazide	5	−40	62
	(pK_a 8.8 and 10.4)	8	−39	59
		9	−42	70
HP-β-CD	Acetylsalicylic acid	1	−68	−166
HP-β-CD	Acetazolamide		−18	−26
HP-β-CD	17β-Estradiol		−71	−151
HP-β-CD	Hydrocortisone		−20	−6
HP-β-CD	Methyl acetylsalicylate	1	−55	−127
HP-β-CD	Methyl salicylate	1	−63	−144
M/DM-β-CD	Acetylsalicylic acid	1	−57	−134
M/DM-β-CD	Methyl acetylsalicylate	1	−20	−28
HP-γ-CD	Acetylsalicylic acid	1	−28	−56
HP-γ-CD	Methyl acetylsalicylate	1	−75	−194
HP-γ-CD	Methyl salicylate	1	−73	−176

[a] HP-α-CD: (2-hydroxypropyl)-α-cyclodextrin . β-CD: β-cyclodextrin. CD: (2-hydroxypropyl)-β-cyclodextrin. M/DM-β-CD: mixture of maltosy dimaltosyl-β-cyclodextrin (3:7). HP-γ-CD: (2-hydroxypropyl)-γ-cyclodextri

enthalpy related to less negative entropy values.[43–45,48] effect, termed compensation, is often correlated with v acting as a driving force in complex formation. The driving force for complex formation could, therefore, b release of enthalpy-rich water from the cyclodextrin cav The water molecules located inside the cavity cannot sa their hydrogen-bonding potentials; therefore, they a higher enthalpy.[51] The energy of the system is lowered these enthalpy-rich water molecules are replaced by sui guest molecules which are less polar than water. C mechanisms that are thought to be involved with com formation have been identified in the case of α-cyclodex In this instance, release of ring strain is thought to be inv with the driving force for compound–cyclodextrin interac Hydrated α-cyclodextrin is associated with an interna drogen bond to an included water molecule which pert the cyclic structure of the macrocycle. Elimination o included water and the associated hydrogen bond is re to a significant release of steric strain decreasing the sy enthalpy.[52] In addition, "nonclassical hydrophobic eff have been invoked to explain complexation. These non sical hydrophobic effects are a composite force in whicl classic hydrophobic effects (characterized by large positiv and van der Waals effects (characterized by negative ΔH negative ΔS) are operating in the same system. U adamantanecarboxylates as probes, α-, β-, and γ-cyclodex were examined.[53] In the case of α-cyclodextrin, experim data indicated small changes in ΔH and ΔS consistent little interaction between the bulky probe and the small ca In the case of β-cyclodextrin, a deep and snug-fitting com was formed leading to a large negative ΔH and a near ΔS. Finally, complexation with γ-cyclodextrin demonstr near zero ΔH values and large positive ΔS values consi with a classical hydrophobic interaction. Evidently, the c size of γ-cyclodextrin was too large to provide for a signif

Table 5—Solubility of Drugs in Different Cyclodextrin Solutions at Room Temperature

Drug	Cyclodextrin[a]	Concn[b] (% w/v)	Solubility (mM)	Enhancement[c] Factor	
Hydrocortisone (MW 362)	None		0.993		
	Glucosyl-α-CD	10	7.45	7.50	
	Maltosyl-α-CD	10	11.3	11.4	
	HP-β-CD MS 0.6	10	33.7	33.9	
	HE-β-CD	10	48.3	48.6	
	RM-β-CD MS 0.6	10	72.2	72.7	
	RM-β-CD MS 1.8	10	50.8	51.2	
	HTMAP-β-CD MS 0.5	10	30.3	30.1	
	CM-β-CD MS 0.6	10	44.6	44.9	
	Glucosyl-β-CD	10	46.9	47.2	
	Maltosyl-β-CD	10	28.7	28.9	
	RM-γ-CD MS 0.6	10	58.8	55.2	
	RM-γ-CD MS 1.8	10	38.6	38.9	
Paclitaxel (Taxol, MW 854)[d]	None		4×10^{-4}		
	β-CD	1.5	0.005	13	
	Dimaltosyl-β-CD	50	0.115	288	
	HE-β-CD	50	0.914	2285	
	HP-β-CD	50	0.856	2140	
	DM-β-CD	50	39.6	99.000	
	γ-CD	15	0.020	50	
	HP-γ-CD	50	0.080	200	
Pancratistatin (MW 325)	None		0.16		
	HTMAP-β-CD MS 1.4	10	0.86	5.4	
	S-β-CD Na-salt MS 2.3	10	0.28	1.8	
	CM-β-CD Na-salt MS 0.6	10	0.83	5.2	
	HP-β-CD MS 0.5	10	1.0	6.3	
	Maltosyl-β-CD MS 0.14	10	0.95	5.9	
	DM-β-CD MS 2.0	10	1.2	7.5	
	HE-β-CD	10	0.83	5.2	
	γ-CD	10	0.80	5.0	
	HTMAP-γ-CD MS 0.3	10	0.49	3.1	
	HP-γ-CD MS 0.7	10	0.83	5.2	
	TM-γ-CD MS 3.0	10	0.49	3.1	

[a] β-CD: β-cyclodextrin. HP-β-CD: (2-hydroxypropyl)-β-cyclodextrin. HE-β-CD: (hydroxyethyl)-β-cyclodextrin. RM-β-CD: randomly methylated β-cyclo HTMAP-β-CD: (2-hydroxy-3-(trimethylammonio)propyl)-β-cyclodextrin. CM-β-CD: (carboxymethyl)-β-cyclodextrin. Glucosyl-β-CD: glucosyl-β-cyclodextrin. M β-CD: maltosyl-β-cyclodextrin. DM-β-CD: 2,6-*O*-dimethyl-β-cyclodextrin. S-β-CD: β-cyclodextrin sulfate. γ-CD: γ-cyclodextrin. RM-γ-CD: randomly meti γ-cyclodextrin. HP-γ-CD: (2-hydroxypropyl)-γ-cyclodextrin. HTMAP-γ-CD: (2-hydroxy-3-(trimethylammonio)propyl)-γ-cyclodextrin. TM-γ-CD: trimethyl γ-cyclo MS: molar substitution (i.e., the average number of OH groups on each glucose repeat unit that have been substituted). Na salt: sodium salt. [b] Concentratior aqueous cyclodextrin solution. [c] The solubility in the aqueous cyclodextrin solution divided by the solubility in water. [d] pH 7.4

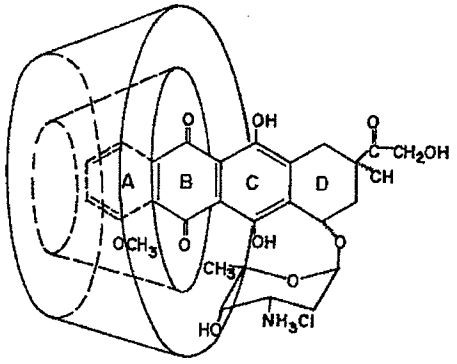
form.[37] The K_c for the phenytoin–β-cyclodextrin complex is over 3 times larger for the un-ionized form than for the anionic form.[46] However, it is frequently possible to enhance cyclodextrin solubilization of ionizable drugs by appropriate pH adjustments. Thus, the solubilizing effects of both (2-hydroxypropyl)-β-cyclodextrin and dimethyl-β-cyclodextrin on dihydroergotamine mesylate have been found to increase with decreasing pH (i.e., formation of the cationic form). Both the saturation solubility and the slopes of the phase–solubility diagrams increase with decreasing pH.[73] Similar results have been reported for the complexation of phenytoin with β-cyclodextrin[46] and for the complexation of indomethacin,[74] prazepam, acetazolamide, and sulfamethoxazole[75] with (2-hydroxypropyl)-β-cyclodextrin.

As mentioned before, it is also possible to enhance complexation and, thus, the solubilizing effect of cyclodextrins by addition of polymers or hydroxy acids to the cyclodextrin solutions. It has been shown that polymers, such as water-soluble cellulose derivatives and other rheological agents, can form complexes with cyclodextrins and that such complexes possess physicochemical properties different from those of individual cyclodextrin molecules.[49,76] In aqueous solutions water-soluble polymers increase the solubilizing effect of cyclodextrins on various hydrophobic drugs by increasing the apparent stability constants of the drug–cyclodextrin complexes. For example, the solubilizing effect of 10% (w/v) (2-hydroxypropyl)-β-cyclodextrin solution on a series of drugs and other compounds was increased from 12 to 129% when 0.25% (w/v) poly(vinylpyrrolidone) was added to the aqueous c dextrin solution.[49] Water-soluble polymers are also ca of increasing aqueous solubilities of the parent cyclodex without decreasing their complexing abilities, thus ma them more feasible as pharmaceutical excipients. Like addition of hydroxy acids, such as citric, malic, or tartaric can enhance the solubilizing effect of cyclodextrins thr formation of super complexes or salts.[67] It is frequ possible to obtain even larger solubilization enhanceme applying several methods simultaneously. For inst prazepam is a benzodiazepine with a pK_a of about 3 Hydroxypropyl)-β-cyclodextrin has a solubilizing effect on the un-ionized and the ionized form of the drug, ar expected, hydroxypropyl methylcellulose has a syner effect on the solubilization. However, the synergistic was more pronounced for the ionized form (Figure Finally, pharmaceutical formulations should contain as an amount of cyclodextrin as possible since excess cyclode can reduce, e.g., drug bioavailability and preservative eff Drug solubility should be determined in the final formul and under normal production conditions to determine much, or too little, cyclodextrin is being used.

Effect on Drug Stability

The effects of cyclodextrins on the chemical stabili drugs is another useful property of these excipients an been extensively examined in the literature.[10] Cyclode

Table 6—Proposed Structure of the Doxorubicin–γ-Cyclodextrin Complex[86] and Stabilization of Doxorubicin and Related Drugs by Cyclodextrin Complexation[87–89]



Drug	pH	Temp (°C)	k_o[a] (min^{-1})	Cyclodextrin[b]	k_c[a] (min^{-1})	k_o/k_c	K_c[a] (M
Daunorubicin	1.5	50	2.16×10^{-3}	M-β-CD	2.64×10^{-4}	8.2	1960
	1.5	50	2.00×10^{-3}	γ-CD	3.72×10^{-4}	5.4	211
Demethoxydaunorubicin	1.5	50	2.16×10^{-3}	M-β-CD	5.40×10^{-4}	4.0	3690
Doxorubicin	1.01	75	0.17	HP-γ-CD	3.02×10^{-2}	5.7	69
	1.84	75	1.86×10^{-2}	HP-γ-CD	2.10×10^{-3}	8.9	193
	5.90	75	1.23×10^{-2}	HP-γ-CD	4.70×10^{-3}	2.6	243
	7.72	75	5.48×10^{-2}	HP-γ-CD	1.03×10^{-2}	5.3	132
	1.5	50	1.71×10^{-3}	γ-CD	3.36×10^{-4}	5.1	197

[a] k_o represents the observed first-order rate constant for the degradation of the free drug, k_c represents the observed first-order rate constant for the degr of the drug within the complex, and K_c is the observed stability constant for the complex, assuming 1:1 complex formation. [b] M-β-CD: methylated β-cyclo γ-CD: γ-cyclodextrin. HP-γ-CD: (2-hydroxypropyl)-γ-cyclodextrin.

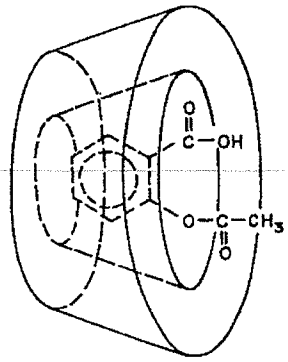
better protection, i.e., a larger stability constant (K_c) and a more favorable k_o:k_c ratio, than γ-cyclodextrin.

Aspirin (acetylsalicylic acid) is a phenolic acetate ester, and thus, it is unstable in aqueous solutions. In acidic buffer solutions (at pH about 1), the ester is hydrolyzed via an $A_{AC}2$ mechanism whereby it undergoes an acyl–oxy cleavage subsequent to protonation, attack by water molecules, and formation of an unstable tetrahedral intermediate.[90] Unionized aspirin forms stable (1:1) inclusion complexes with the various β-cyclodextrins. NMR studies have shown that in the complex the benzene ring is located well inside the cavity with the acetyl ester group protruding from cavity. This location of the acetyl ester does not completely prevent its hydrolysis but due to steric hindrance, the hydrolysis was determined to be 4–6 times slower within the complex than outside it (i.e., the k_o:k_c ratio in Table 7 is between 4 and 6). However, under neutral conditions, where aspirin is in the ionized form, the same cyclodextrins did not affect the observed hydrolytic rate constant. NMR studies indicated that the ionized aspirin does not form complexes with the β-cyclodextrins tested. The cyclodextrins did not influence the kinetic behavior (e.g., the order of reaction) or the degradation mechanism, only the rate of reaction.[48]

Sulfobutyl ether β-cyclodextrin, which is an anionic β-cyclodextrin derivative, has been shown to be highly effective in improving the chemical stability of the antitumor drug O^6-benzylguanine. The benzyl moiety of the drug was responsible for the cyclodextrin complex formation resulting in an objective increased shelf-life of an aqueous parenteral O^6-benzylguanine formulation.[71] The same cyclodextrin derivative has been used to increase the shelf-life (and ocular absorption) of pilocarpine in aqueous eye drop solutions.[91] The cyclodextrin stabilization of pilocarpine appeared to be independent of the drug ionization status. Another anionic type cyclodextrin, i.e., O-(carboxymethyl)-O-ethyl-β-cyclodextrin, has been used to stabilize prostaglandin E_1 in a fatty alcohol propylene glycol ointment.[92] Dihydroergotamine nasal spray has used as an acute treatment of migraine. However, dih ergotamine, the free base, has both limited aqueous solu and stability. Cyclodextrins, such as (2-hydroxyprop cyclodextrin, have been used to solubilize the drug in aqu solutions and to stabilize it during autoclaving.[73]

Degradation kinetics in the solid state are, in general, complicated and they progress more slowly than in aqu solutions. Consequently, there are fewer reports on the e of cyclodextrins on the solid-state decomposition of d

Table 7—Proposed Structure of the Aspirin–β-Cyclodextrin Comple Stabilization of Aspirin by Cyclodextrin Complexation[48]



Drug	pH	Temp (°C)	k_o (min^{-1})	Cyclodextrin[a]	k_c (min^{-1})	k_o/k_c	K
Aspirin	Ca. 1	65	4.76×10^{-3}	H-β-CD	1.11×10^{-3}	4.3	
				M/DM-β-CD	8.25×10^{-4}	5.8	
				HP-γ-CD	1.18×10^{-3}	4.0	

[a] HP-β-CD: (2-hydroxypropyl)-β-cyclodextrin. M/DM-β-CD: mixture of m and dimaltosyl-β-cyclodextrin (3:7). HP-γ-CD: (2-hydroxypropyl)-γ-cyclo

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